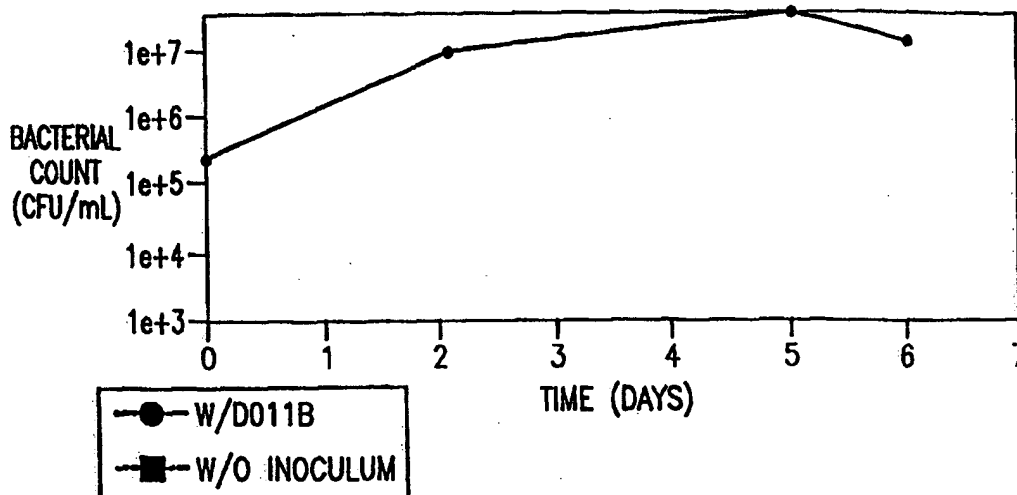




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(54) Title: BIOLOGICAL DEODORIZING LIQUID COMPOSITION



## (57) Abstract

The composition of the invention comprises an aqueous mixture of an odor neutralizer component, an enhancer component for microbial activity, and a microbial component. This composition is designed to provide short- and long-term odor control effects and is environmentally friendly and economical for use.

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## BIOLOGICAL DEODORIZING LIQUID COMPOSITION

### Cross Reference to Related Application

Reference is made to and priority claimed from U.S. Provisional Application Serial No. 60/077,544 filed March 11, 1998, entitled BIOLOGICAL DEODORIZING LIQUID COMPOSITION.

### 5      **Field of the Invention**

This invention is directed to a novel biological deodorizing liquid composition which is designed to be applied in the areas of pet care, toilet care, carpet care, and garbage collections or processes, management of industrial wastes, including sludge processing, landfill and composting, and odor control of livestock  
10      production processes and other organic wastes.

### **Background of the Invention**

Offensive odors are generated from various sources, including pet wastes, toilets, carpets, garbage collections and processes, animal manure, industrial waste sites such as sludge processes, landfill sites, and composting sites, etc. Among the  
15      odorous compounds, amines, ammonia, hydrogen sulfide, organic acids, and mercaptans are very often found in the malodors from various sources and they are, respectively, the products of decomposition and other reactions of organics and nitrogen- and sulfur-containing materials. Offensive odors have posed a series of social and environmental problems including hazards to mental health, damages to  
20      health of humans, especially the workers in odor-generating facilities, and negative effects on animal growth and reproduction.

Conventionally, masking agents, such as fragrances, have been used to cover  
up an objectionable odor with a more desirable one. However, masking agents may not actually reduce concentrations of odorous gases and they also quickly lose their  
25      effectiveness due to vaporization and microbial break down. Chemical oxidizing agents and germicides have also been used to control odors by altering or eliminating bacterial action responsible for odor production. These chemical agents,

however, will destroy the beneficial microbial activity in the treated systems. Furthermore, some of them might not be safe for humans and animals and are usually expensive for use. Other deodorizing approaches include use of adsorbents, neutralizers, and biological degradation or conversion. Adsorbents are products with a large surface area that may be used to adsorb the odors before they are released to the environment. Neutralizers are materials which react with odorous compounds to form odorless ones. Biological degradation or conversion can eliminate odors through biochemical digestive processes. The biological approaches include: 1) use of externally added microbes and enzymes; and 2) use of enhancing agents to ensure or increase the activity of added microbes and indigenous microbial populations.

Although the above technical concepts have been known for some time, how to use these concepts more efficiently and how to improve these concepts for development of new deodorizing products have long been in demand. The requirements for the new deodorizing products might include: 1) products have short-term and long-term deodorizing efficacy; 2) products can enhance beneficial microbial activity; and 3) products are environmentally friendly and economical. It is obvious that use of masking agents, chemical oxidizing agents, and germicides might not meet these requirements.

Use of a biological approach is a promising one, since it can eliminate odors through biodegrading odor sources including organics and nitrogen- and sulfur-containing materials, thus providing long-term odor control. This approach is environmentally friendly and usually economical. Because odorous compounds are very volatile, rapid containment of odors, through using adsorbents and/or neutralizers, is usually necessary before the odors are released to the environment. This rapid containment action can provide short-term odor control and retain the odorous compounds for the subsequent biodegradation when beneficial microbial activity is maintained in the treated system. Therefore, use of a biological approach in conjunction with neutralizers should be a reasonable option for providing short- and long-term odor control. The technical challenges for realizing this option include: 1) how to develop a stable liquid composition that contains neutralizers, a

microbial activity enhancer, and microbes; 2) how to identify suitable neutralizers compatible with the other components of the composition; and 3) how to select suitable ingredients and microbes that enhance and provide the beneficial microbial activity.

5           The objective of this invention was to provide a stable liquid deodorizing composition that contains neutralizers, a microbial activity enhancer, and microbial strains.

          Hata, U.S. Patent 4,879,238, teaches the deodorization by using a single strain or a few strains of bacteria. Further, Kurasawa, U.S. Patent 4,996,055,  
10       presents a deodorant that contains genus butyric acid bacteria and *Bacillus subtilis* as effective components for treating excrement of various animals and other sources of foul odors. The prior art suggests that use of specially-selected microorganisms can reduce malodors. However, none of the foregoing art teaches the use of a combination of an odor neutralizer, a microbial activity enhancer, and microbes for  
15       short- and long-term odor control.

### Summary of the Invention

          This invention provides a liquid composition comprising a mixture of an odor neutralizer component, an enhancer component for microbial activity, and a microbial component. This composition has advantages of being an effective odor  
20       control agent, a good enhancing agent for microbial activity, and a microbial additive for degradation of odorous sources. This composition represents a novel concept that provides short- and long-term odor control effects in an environmentally friendly and economical manner.

          Another feature of the present invention is that a unique odor neutralizing chemical, propylene carbonate, is selected. In combination with other components  
25       of the composition, propylene carbonate can effectively reduce odors, including amine and ammonia odors.

          An additional novel aspect of this invention is that a plant extract, yucca schidigera, has been found to be effective for enhancing activity of microorganisms,

both introduced and indigenous. Our tests have demonstrated that use of yucca schidigera, in conjunction with microbial strains and other ingredients of the formulations, can significantly enhance the growth of microbes in the treated system, thus increasing the biodegradation of odor sources.

5 Furthermore, a series of microbial strains have been selected to be compatible with the composition of the invention. These microbes can seed the treated system and start developing a beneficial microbial population.

### **Brief Description of the Drawing**

10 For a fuller understanding of the nature and objects of the invention, reference should be made to the following detailed description of a preferred mode of practicing the invention, read in connection with the accompanying drawings, in which:

FIG. 1 illustrates a plot of microbial growth and biodegradation of ammonia and COD by DO11B.

### **Detailed Description of the Invention**

15 The composition of the invention comprises a liquid mixture of an odor neutralizer component, an enhancer component for microbial activity, and a microbial component. This composition is designed to provide short- and long-term odor control effects and is environmentally friendly and economical for use.

20 The composition of the invention does not rely on fragrances for odor control. Instead, the composition utilizes unique neutralizer materials to rapidly decrease odorous compounds by converting them to delete odorless ones. Moreover, the composition of the invention provides two novel mechanisms for enhancing microbial activity, thus resulting in effective biodegradation of odor sources  
25 including organics and nitrogen- and sulfur-containing wastes. The mechanisms for enhancing microbial activity provided by this invention include the use of a unique microbial activity enhancing component and the use of a blend of specially-selected microbial strains. The novel combination of odor neutralizing, microbial activity

enhancing, and microbial components makes the composition of the invention the one that not only can rapidly reduce malodor but also can biologically reduce or eliminate odor sources in a longer term.

#### Odor Neutralizer Component

5 Odor neutralizers are the agents that can rapidly interact, by chemical reactions, with odorous compounds to produce odorless compounds. These agents should not rely on the masking mechanism of a perfume to control odors. In addition, these agents must be safe for use and cost effective. Neutralizers used in the invention must be compatible with the microbial activity enhancer and  
10 mircrobial components.

In one embodiment of the present invention, a unique chemical neutralizer, propylene carbonate, was found. The molecular formula of propylene carbonate is  $C_4H_6O_3$ . A preferred product of propylene carbonate is available from commercial vendors such as Huntsman Chemical Corporation. The following experiment was  
15 carried out to examine the effectiveness and the required dosage rate of propylene carbonate for odor control.

Two prototypes, one containing propylene carbonate and the other containing no propylene carbonate, were prepared as shown in Example 1 and Table 4. To each 25-ml test tube, 9 ml of trimethylamine (0.1% w/v), dimethylamine (0.15% w/v), or  
20 ammonia (1% w/v ammonium chloride, pH 9.5) solution was added. Each sample was then dosed, respectively, with 0, 1.5, and 2.5 ml of the prototype with or without propylene carbonate. The test tubes were capped, vortexed for 10 seconds, and then left stationary at 23 °C. After 2 hrs, Gastec detector tubes (Gastec Corporation) were used to determine trimethylamine, dimethylamine, and ammonia  
25 concentrations, respectively, in the head space of the test tubes. As shown in Table 1, the prototype containing propylene carbonate significantly outperformed the prototype without propylene carbonate in reduction of all three odorous compounds. The test results also provide information about the dosage rate of the prototype required to treat a given amount of odorous compound. These results have

demonstrated that a suitable dosage of propylene carbonate, combined with other components of the formulation, can effectively reduce the odors of trimethylamine, dimethylamine, and ammonia, which are the major target odorous compounds. In addition, propylene carbonate does not inactivate the microbial activity enhancer and the microbial components even after a long period of contact. Toxicity tests have also shown that propylene carbonate is safe for the applications of the invention. Propylene carbonate, in combination with other components of the formulation, functions to provide a unique composition unavailable in the art.

**Table 1. Effect of Propylene Carbonate on Reduction of Odorous Compounds**  
(Head space gas measured by gas detector; treatment time: 2 hrs)

Odorous compound	% odor reduction by different volume of prototypes with and without propylene carbonate (PC)				
	0 ml prototype (control)	1 ml prototype		2.5 ml prototype	
		w/PC	w/o PC	w/PC	w/o PC
Trimethylamine	0	96.8	32.3	97.8	35.0
Dimethylamine	0	87.2	30.9	89.7	29.9
Ammonia	0	48.6	20.0	80.0	24.0

Other odor neutralizing compounds, such as sodium citrate, sodium bicarbonate, and sodium carbonate, may also be used in the formulation of this invention.

#### Microbial Activity Enhancer Component

A microbial activity enhancer is defined as an agent that can enhance growth of microbial populations, indigenous and/or introduced, in a treated system, thus increasing the microbial activity for degradation of odor sources. The activity enhancer must be safe for use and cost effective.

This invention has found that yucca schidigera, combined with other components, is a very effective enhancer for desired microbial activity. Yucca



shidigera is a natural plant extract and has a long history of safe use as a food material for both humans and livestock. The combination of yucca schidigera with odor neutralizing and microbial components has formed another unique aspect of the invention.

5 A preferred liquid product of yucca schidigera (e.g., available from commercial vendors such as Desert King International Corporation) should have the following properties: active ingredient, 99.9% yucca schidigera extract; density, 10 lbs/gallon; and pH, 3.6 - 4.0.

10 The following experiment was conducted to examine the effect of yucca schidigera on microbial activity. One hundred ml of culture medium containing mineral salts and 0.2% (w/v) glucose was added into each 250-ml flask. Four *Bacillus* strains, *Bacillus licheniformis* culture DA33 (ATCC 55406), *Bacillus amyloliquefaciens* culture SB 1002 (Sybron Chemicals Inc.), *Bacillus pasteurii* culture SB 1003 (Sybron Chemicals Inc.), and *Bacillus laevolacticus* culture SB 15 1006 (Sybron Chemicals Inc.), were used to inoculate the media. Yucca schidigera (0.02 ml of DK50 from Desert King International Corporation) was added to one set of the samples while the other set of samples received no yucca schidigera. The flasks were placed on a shaker at 23 °C and the optical density (proportional to bacterial concentration) of the samples was measured over time by a 20 spectrophotometer set at 580 nm. The bacteria in the samples containing yucca schidigera grew much better than in the samples without yucca schidigera, as indicated by the change in optical density (Table 2). This result demonstrates that yucca schidigera can significantly enhance growth of a specially-selected microbial blend.

25 **Table 2. Effect of Yucca Schidigera (YS) on Microbial Growth**

Treatment	<u>Microbial concentration in media (OD<sub>580</sub>) at different time</u>		
	0 hr	102 hrs	174 hrs
without YS	0.013	0.025	0.020
with YS	0.022	0.29	0.92

### Microbial Component

5 Viable microorganisms, or mixture thereof, which are capable of growing on and degrading common domestic, industrial, pet, and animal wastes, capable of surviving the formulations, and compatible with the formulations, and do not produce malodor while performing, may be used in the invention.

The invention has found that suitable types of microorganisms include *Bacillus* strains. *Bacillus* strains are spore formers. They are stable in the formulation and can function to degrade odor sources when the formulation is applied.

### 10 Other Ingredients

Depending on the applications, other ingredients used in the invention include surfactant, fragrance, and dye.

15 Surfactants can wet and emulsify insoluble waste materials present in the treated system and inclusion of surfactants in the composition of the invention will add to it a cleaning capability. Furthermore, surfactants can be used to break down the insoluble wastes therefore increasing the availability of them to microbial degradation. Suitable surfactants for the invention include nonionic and anionic types.

20 Fragrance and dye can be optionally added to mask the odor and to control the color of the composition of the invention, respectively, and for market appeal. The fragrance and dye must be compatible with other ingredients of the composition.

The composition in this invention can typically have the following ingredients (Table 3).

**Table 3. Typical Formulation of the Invention**

Components	Broad	Preferred
Odor neutralizers	1-15%w	2-10%w
Microbial activity enhancer	0.2-5%w	1-3%w
Microbes	$1 \times 10^5$ - $1 \times 10^{10}$ /ml	$1 \times 10^6$ - $1 \times 10^8$ /ml
Surfactants	0-8%w	0-6%w
Preservatives	0.02-0.2%w	0.04-0.1%w
Fragrance	as needed	as needed
Dye	as needed	as needed
Water	Balance	Balance

The invention is further illustrated by the following examples which constitute preferred embodiments of the invention.

**Example 1**

A liquid formula of DO11B was prepared as shown in Table 4. An odor neutralizer (propylene carbonate), a microbial activity enhancer (yucca schidigera), a blend of bacterial spores, surfactants, nutrients, and preservatives were used in this formula. Each ingredient was added to water and mixed to form a stable aqueous solution. DO11B is designed to be sprayed on the odor-generating wastes for deodorizing amines, ammonia, and other odorous compounds. Its applications include pet care, toilet care, carpet care, garbage collections or processes, landfill, composting, etc.

Table 4. DO11B Formulation

Ingredient	Concentration		
5	Propylene carbonate	5.98%w	
	Yucca schidigera	1.04%w	
	<i>Bacillus</i> strains	4x10 <sup>7</sup> /ml	
	including: <i>Bacillus licheniformis</i>	culture DA33*	70% by count
	<i>Bacillus amyloliquefaciens</i>	culture SB1002*	10% by count
10	<i>Bacillus Pasteurii</i>	culture SB1003*	10% by count
	<i>Bacillus laevolacticus</i>	culture SB1006*	10% by count
	Ethoxylated alcohol	0.68%w	
	Linear alkyl benzene sulfonic acid	0.80%w	
	Sodium nitrate	0.19%w	
15	Dipotassium phosphate	0.39%w	
	Methyl anthranilate	0.0062%w	
	5-Chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-4-isothiazolin-3-one	0.024%w	
	1,2 Benzisothiazolin-3-one	0.0057%w	
20	EDTA	0.079%w	
	Water	balance	
	pH	6.3-7	

\*Microbial strains produced by Sybron Chemicals Inc.

25 In a test, DO11B was used to treat three aqueous solutions of trimethylamine, dimethylamine, and ammonia, respectively, contained in capped test tubes (test procedure is the same as listed in the previous section of this patent). After a 2-hr treatment, a gas sample was taken from the head space of each test tube and a gas detector was used to measure the concentration of each odorous compound in the gas sample. As shown in Table 5, DO11B significantly reduced the concentrations of trimethylamine, dimethylamine, and ammonia in the gas phase.

Another laboratory test was conducted to examine the effect of microorganisms on odor control. A synthetic sludge was prepared, which contained

-11-

corn starch (0.6% w/v), skim milk (1.0% w/v), gelatin (0.2% w/v), beef extract (0.78% w/v), Crisco vegetable shortening (0.2% w/v), dextrose (1.0% w/v), skin lotion (0.2% w/v), Sunlight dish detergent (0.2% w/v), baking soda (0.1% w/v), sodium chloride (0.04% w/v), and urea (0.045% w/v) in water. Two ml of this synthetic sludge and 5 ml of 2% (w/v) solution of ammonium chloride were added to 100 ml of distilled water contained in a 250-ml flask. To a set of the samples, 0.1 ml of DO11B was added as inoculum while the other set received no DO11B. The flasks were placed on a shaker at 23 °C and the total microbial count (measured by standard method agar), soluble COD (chemical oxygen demand; measured by HACH method) concentration, and ammonia concentration (measured by Phenate method) were monitored over time. As shown in Figure 1, after the addition of DO11B to the synthetic sludge medium, the bacteria in DO11B grew and biologically reduced ammonia and COD, thus reducing or eliminating the odor sources.

These results have demonstrated the efficacy of DO11B for odor control and the benefits of using microorganisms in the odor control formula.

**Table 5. Reduction of Amines and Ammonia by DO11B**

**(Head space gas measured by gas detectors;**

**9 ml odorous solution; treatment time: 2 hrs)**

Odorous compound	<u>Odor conc. (ppm) after treatment w/different vol. of DO11B</u>					
	0 ml	0.5 ml	1 ml	1.5 ml	2 ml	2.5 ml
Trimethylamine	>600	44	20			
Dimethylamine	150	10				
Ammonia	540	260	140	90	70	60

**Example 2**

Another formula, DO11A, was prepared and tested to deodorize two landfill waste materials, a sludge and a press cake, from a paper mill. All the active ingredients in DO11B were contained in DO11A along with additional 0.2% of herbal fragrance. The use of the fragrance was mainly for market appeal. Twenty grams of each landfill sample was added into a 100-ml beaker. Each sample was dosed with 1:2 diluted (1 part DO11A diluted with 1 part water) and undiluted DO11A. All the test beakers were then covered with parafilm. Gastec detector tubes (Gastec Corporation) were used to determine amine and ammonia concentrations in the head space of the beakers. A panel was also asked to smell the samples. At 4.5 hrs and 69.5 hrs after applying DO11A (Tables 6 and 7), amine and ammonia emitted from the two landfill samples were significantly decreased, resulting in a significant odor reduction as determined by a panel.

**Table 6. Odor Reduction of a Landfill Sample by DO11A**  
(20 g of landfill sample used in the test)

Treatment	<u>Amine (ppm)</u>		<u>Ammonia (ppm)</u>		<u>Odor Strength</u>
	4.5 hr	69.5 hr	4.5 hr	69.5 hr	(Ranking)*
Not treated	616	240	100	45	5
w/1 ml 1:2 DO11A	NM**	200	87	30	3.5
w/1 ml DO11A	NM	224	80	28	2
w/2 ml DO11A	280	200	60	28	0.5

\*Odor strength was determined by a panel; 5 indicates strongest odor in the test, 0 indicates no odor.

\*\*NM: Not measured.

**Table 7. Odor Reduction of a Landfill Press Sample by DO11A  
(20 g of landfill press sample used in the test)**

Treatment	Amine (ppm)	Odor Strength (Ranking)*
	4.5 hr	69.5 hr
Not treated	10	5
w/1 ml 1:2 DO11A	0	4.5
w/1 ml DO11A	0	3.5
w/2 ml DO11A	0	2.5

\*Odor strength was determined by a panel; 5 indicates strongest odor in the test, 0 indicates no odor.

While specific embodiments of the invention have been illustrated and described herein, it is realized that modifications and changes will occur to those skilled in the art. It is therefore to be understood that the appended claims are intended to cover all modifications and changes as fall within the true spirit and scope of the invention.

**We claim:**

- 1 1. A liquid biological deodorizing composition which comprises:
  - 2 an odor neutralizing component which functions to provide for rapid odor
  - 3 reduction;
  - 4 a microbial activity enhancing component which provides for the
  - 5 acceleration of microbial growth and the biodegradation of odor sources;
  - 6 a microbial blend that can seed the treated systems to stimulate
  - 7 biodegradation of odor sources;
  - 8 with said components all being contained in a stable aqueous medium, and
  - 9 wherein said odor neutralizing component, microbial activity enhancing component,
  - 10 and microbial blend are present in concentrations sufficient to control odor and to
  - 11 induce beneficial microbial degradation of odor sources.
- 1 2. The composition of claim 1 in which the odor neutralizing component comprises
  - 2 propylene carbonate.
- 1 3. The composition of claim 1 in which the odor neutralizing component is at least
  - 2 one selected from the group consisting of sodium citrate, sodium bicarbonate, and
  - 3 sodium carbonate.
- 1 4. The composition of claim 1 in which the microbial blend comprises at least two
  - 2 *Bacillus* strains.
- 1 5. The composition of claim 4 in which the microbial strains are present in a
  - 2 concentration of from about  $1 \times 10^5$  to  $1 \times 10^{10}$  per ml.



- 1 6. A deodorizing composition in the form of an aqueous solution which  
2 comprises an odor neutralizing material, and an enhancer for microbial activity, and  
3 selected microbes for the degradation of odorous sources, with said components  
4 being present in the following concentrations:

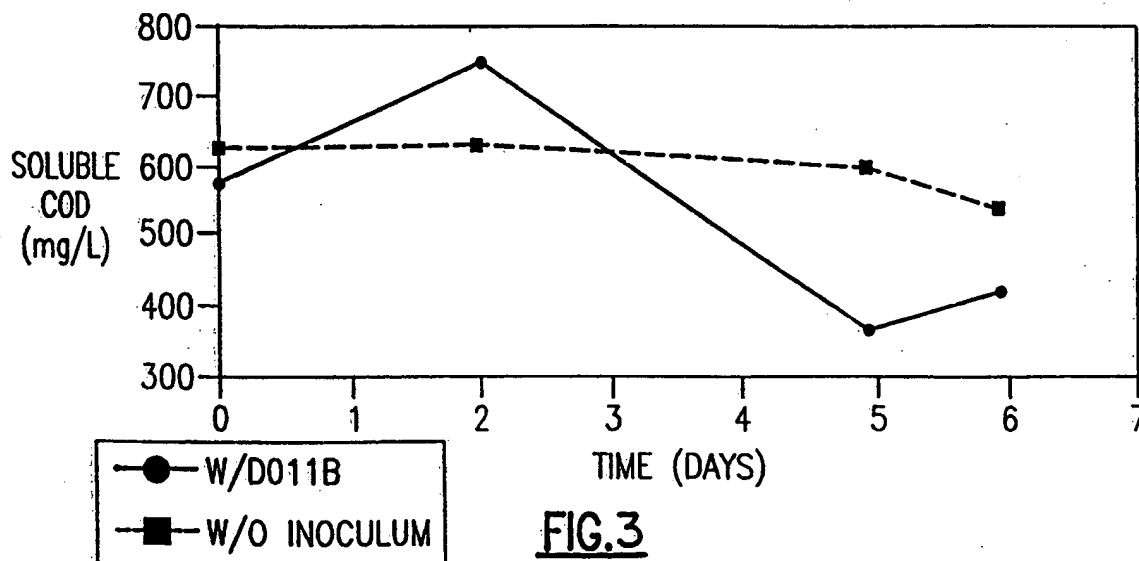
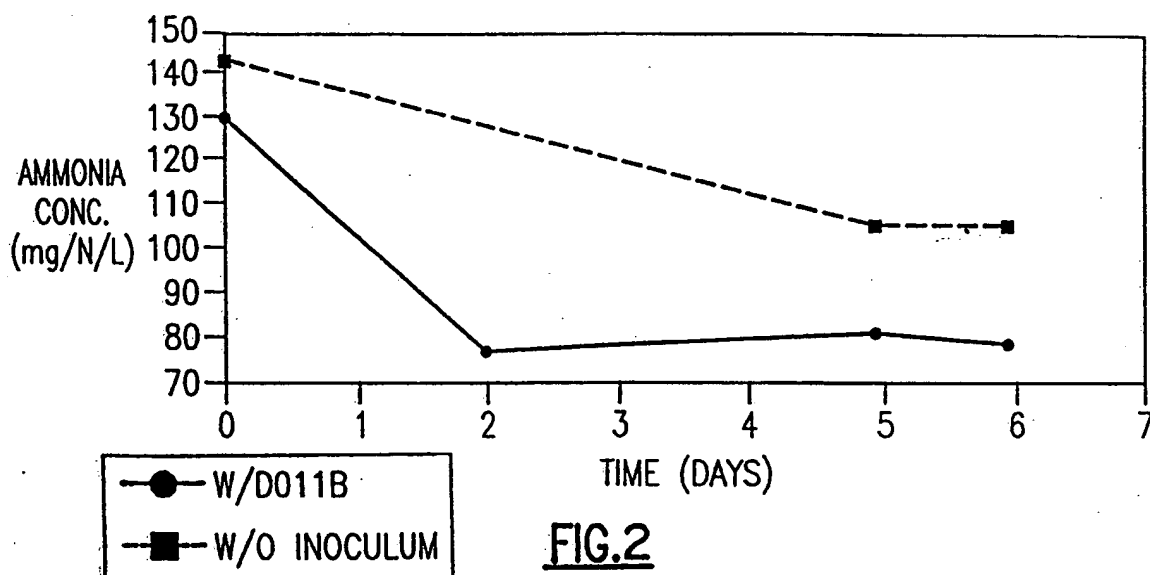
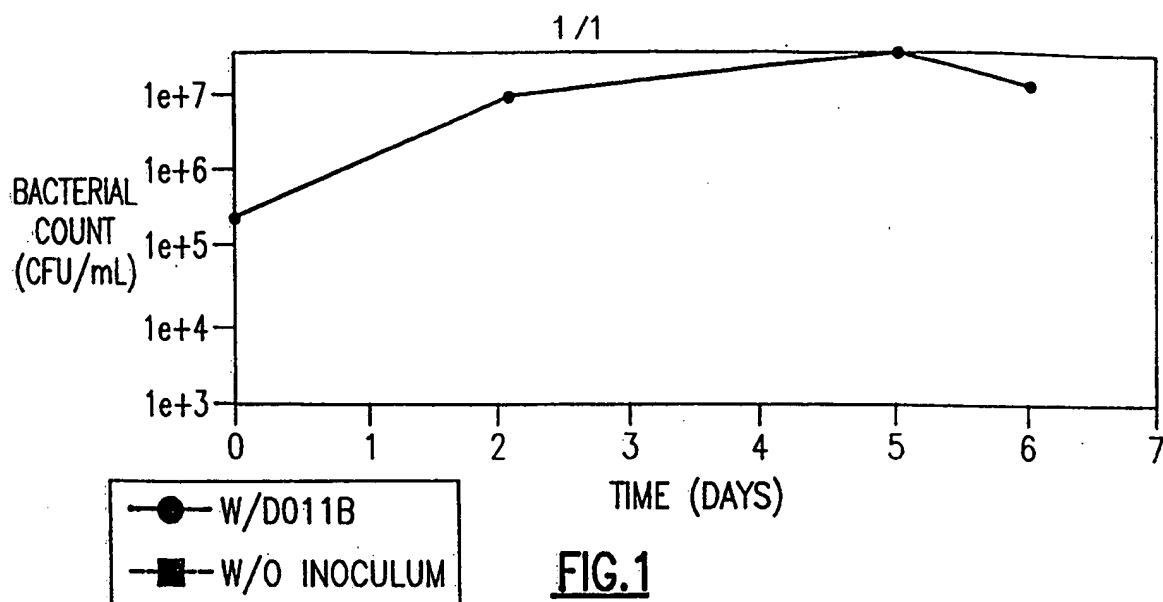
5	<u>Component</u>	<u>Concentration</u>
6	Odor neutralizing material	1-15% wt.
7	Microbial activity enhancer	0.2-5.0% wt.
8	Microbes	$1 \times 10^5$ - $1 \times 10^{10}$ /ml
9	Water	Balance

- 1 7. The composition of claim 6 in which the components are present in the  
2 following concentrations:

3	<u>Component</u>	<u>Concentration</u>
4	Odor neutralizing material	2-10% wt.
5	Microbial activity enhancer	1-3% wt.
6	Microbes	$1 \times 10^6$ - $1 \times 10^8$ /ml
7	Water	Balance

- 1 8. The composition of claim 6 in which the odor neutralizing material  
2 comprises propylene carbonate, the microbial enhancer includes yucca schidigera,  
3 and the microbes include a blend of *Bacillus* strains.

- 1     9.     The composition of claim 1 in which the odor neutralizer comprises  
2     propylene carbonate and the microbes include a blend of at least two selected from  
3     the group consisting of:  
4     *Bacillus licheniformis*  
5     *Bacillus amyloliquefaciens*  
6     *Bacillus pasteurii*  
7     *Bacillus laevolacticus*



## INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US99/04812

## A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) : C09K 3/00; A01N 43/08; A61K 31/34

US CL : 252/380, 382, 384; 514/762; 424/76.1, 76.5, 76.8

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 252/380, 382, 384; 514/762; 424/76.1, 76.5, 76.8

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

APS: odor, microbial, propylene carbonate

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 4,879,238 A [HATA] 07 NOVEMBER 1989, col. 1, lines 10-45.	1-9
Y	US 4,996,055 A [KURASAWA] 26 FEBRUARY 1991, col. 2, lines 48-68.	1-9
Y	US 5,122,301 A [MCCOY et al] 16 JUNE 1992, col. 2, lines 29-47.	1-9



Further documents are listed in the continuation of Box C.



See patent family annex.

* Special categories of cited documents:	*T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
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Date of mailing of the international search report

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